II. REMARKS

Upon entry of the present amendment, claims 1 to 3, 5, 13, 14, 20, and 40 to 58 will be pending.

Applicants and Applicants' representative gratefully acknowledge the Examiner's consideration of the Response filed May 13, 2004, and the clarification of the remaining issues in the Advisory Action mailed July 9, 2004. The amendments and following remarks are intended to address the outstanding issues, including the nature of the expressed truncated Activin Type II receptor (Act RII) and level of expression required for the dominant negative effect.

A. Regarding the Amendments

Claims 1, 13, 20 and 40 to 42 have been amended to clarify that the "truncated" Activin Type II receptor is a truncated "dominant negative" receptor. The amendment is supported, for example, at paragraph 349 (page 128), and, therefore, does not add new matter.

Claims 1, 2 13, and 40 to 42 have been amended to clarify that a "regulatory element comprising a muscle-specific promoter" is used in the constructs. The amendment provides for consistent claim language, which previously recited, for example, a "muscle specific promoter" (claim 1), wherein the "promoter is a ... promoter/enhancer" (claim 2), and a "muscle-specific control sequence" (claim 13); and is supported, for example, at paragraph 101 (pages 37-38), and paragraph 291 (page 105). As such, it is submitted that the amendment does not add new matter.

Claims 2 and 14 have been amended to clarify that the regulatory element is a "myosin light chain promoter and an enhancer". The amendment is necessitated to avoid a potential ambiguity, wherein the term "myosin light chain promoter/enhancer" could be interpreted to mean a "myosin light chain promoter and [any] enhancer" or a "myosin light chain promoter and

1/3 enhancer". The amendment to claims 2 and 14 is supported, for example, at paragraph 99 (page 37), paragraph 101 (pages 37-38), paragraph 144 (page 55), and paragraph 191 (page 72), and, therefore, does not add new matter.

New claims 46 to 58 have been added. New claims 46 and 47, which depend from claims 1 and 13, respectively, specify that the regulatory element comprises "a myosin light chain promoter and 1/3 enhancer". New claims 46 and 47 are supported, for example, at page 126, paragraph 345, and, therefore, do not add new matter.

New claims 48 to 58 are based, in part, on previously pending claims 1, 3, 5, 43, 20, 13, 40, 44, 41, 45, and 42, respectively, and further specify that the truncated Activin Type II receptor is a "dominant negative" Act RII, and that the regulatory element comprises a "myosin light chain promoter and 1/3 enhancer". As such, new claims 48 to 58 are supported by the previously pending claims (which, in turn, are supported for reasons of record), and, for example, at paragraph 345 (page 126) and paragraph 349 (page 128). Accordingly, it is submitted that claims 48 to 58 do not introduce new matter.

B. The Invention

The present invention is based, in part, on Applicants' discovery that Activin Type II receptors (Act RII) specifically bind and mediate signal transduction of myostatin (GDF-8), which is a negative regulator of muscle cell growth. The invention is based further on Applicants' demonstration that expression of a dominant negative Act RII, which lacks kinase activity, in muscle cells inhibited the negative regulatory effect of myostatin, resulting in increased muscle mass in transgenic animals expressing the dominant negative Act RII (see, e.g., paragraph 84 (page 31), paragraphs 111-112 (pages 42-43), and paragraph 119-120 (page 45)). The specification discloses that expression of a dominant negative Act RIIB causes increased

muscle mass in a transgenic animal similar to that observed in myostatin knockout mice (see Example 15; see, e.g., paragraph 351, page 129). More specifically, the specification discloses that seven founder animals were identified that tested positive for the truncated Act RIIB transgene, and that all seven exhibited increased skeletal muscle mass (see page 128, paragraph 349; and Table 2, page 133 - "dom. neg. Act RIIB", males and females). Further, while muscle weights varied among the founder animals, the magnitude of the increase was highly consistent in offspring of the founders (see page 128, paragraph 350; and Table 3, page 134).

C. Rejections under 35 U.S.C. § 112

In the Final Office Action mailed January 13, 2004, the pending claims were rejected under 35 U.S.C. § 112, first paragraph, as allegedly lacking enablement. In the Advisory Action mailed July 9, 2004, it was stated that the rejection was maintained because the claims encompass sequences that do not encode a dominant negative Act RII, and because empirical experimentation would have been required to determine an adequate level of expression of the dominant negative Act RII to compete with endogenous Act RI such that an observable phenotypic effect would occur.

Dominant Negative Act RII

In a telephone conference, the Examiner noted that the recitation of "a truncated Activin Type II receptor...which lacks kinase activity" encompasses a large number of compositions, some of which can be as small as a few amino acids, and that many of the compositions would not have dominant negative Act RII activity. The claims have been amended to clarify that the constructs recited in the claims encode a truncated "dominant negative" Act RII. In view of this amendment, and for the reasons of record in this case, it is submitted that one skilled in the art would have known how to make and use a truncated dominant negative Act RII for practicing

the claimed invention. Accordingly, it is respectfully requested that this basis of the rejection be removed.

Level of Expression of Dominant Negative Act RII

The remaining issue relates to a level of expression of the dominant negative Act RII required to obtain a phenotypic effect (i.e., increased muscle mass). It was acknowledged in the previous Office Action that expression from the myosin light chain promoter and 1/3 enhancer provides sufficient dominant negative Act RII expression for increased muscle mass to be evident, but maintained in the Advisory Action that the claims are not enabled for the use of any muscle specific promoter because it is not predictable that expression of the dominant negative Act RII would be sufficient to obtain the phenotypic effect.

As an initial matter, Applicants point out that new claims 48 to 58 require the myosin light chain promoter and 1/3 enhancer. As such, it is submitted that claims 48 to 58 are not subject to this basis of the rejection.

With respect to the remaining pending claims, Applicants maintain that essentially any level of expression of the dominant negative Act RII in muscle would result in at least some increase in muscle mass. For the reasons of record and as set forth below, it is maintained that one skilled in the art, viewing the subject application, and having knowledge of the art, reasonably would have predicted that any level of expression of a dominant negative Act RII would be expected to result in some increase in muscle mass of the transgenic mammals.

First, as discussed in Applicants' Response filed May 13, 2004, the specification discloses that, following pronuclear injection of the exemplified transgene, which comprises the myosin light chain promoter and 1/3 enhancer, seven founder animals were identified that tested positive

for the truncated Act RIIB transgene, and all seven exhibited an increase in skeletal muscle mass as compared to controls (see page 128, paragraph 349; and Table 2, page 133 - panels labeled "dom. neg. Act RIIB", males and females). As is evident from Table 2, the amount of increased muscle mass varied in the different founder animals, though each shows increased muscle mass. Although not suggesting a mechanism by which variable muscle mass increase occurred, one explanation could be that the transgene integrated in different positions in the genomes of the different founder animals, and that variable expression of the transgene occurred due to the well known positional effects that occur depending on the chromosomal location of the transgene integration. Assuming such positional effects occurred, the results in Table 2 (page 133) indicate that, regardless of where in the genome a transgene may insert, the encoded truncated dominant negative Act RII was expressed at some level, resulting in increased muscle mass in the transgenic animals. Further, offspring of each founder animals exhibited consistent increases in muscle mass; for example, the muscles of male and female offspring from the C5 founder weighed 30-60% more than controls, and the muscles of male and female offspring of the C11 founder weighed 110-180% more than controls (page 128, paragraph 350; and Table 3, page 134). These results demonstrate that, even if different levels of the dominant negative Act RII are expressed due, for example, to a position effect in the genome, the transgenic mammals nevertheless will exhibit a measurable increase in muscle mass, and will be able to transfer the phenotype to their offspring.

Second, Applicants point to the Tsuchida et al. reference, which was submitted as Exhibit D with the Response filed July 3, 2003. For the Examiner's convenience, a copy of Tsuchida et al. is attached hereto as Exhibit B. Tsuchida et al. report that expression of a reporter gene was inhibited in a dose-dependent manner by a dominant negative Act RII (see page 5498, right column, first full paragraph; see, also, Figs. 5A and 5B, page 5500). Briefly, Tsuchida et al. expressed a luciferase reporter gene containing a TPA responsive element in

CHO cells (see page 5495, left column, second full paragraph), and demonstrated that expression of the reporter gene is induced by activin, and that co-expression of a dominant negative Act RII inhibits the activin-induced expression of the reporter gene in a dose-dependent manner. Applicants submit that the skilled artisan, viewing the subject application, and having knowledge of the results of Tsuchida et al., reasonably would have predicted that expression of a dominant negative Act RII would inhibit GDF-8 (myostatin) induced signal transduction in a dose-dependent manner, and that the phenotype due to GDF-8 similarly would be affected in a dose-dependent manner.

Third, Applicants submit that the skilled artisan also would have known that induction of an immune response specific for GDF-8 (myostatin), using various immunogenic fragments of GDF-8, result in increased muscle mass in mammals. For example, WO 99/42573, a copy of which is attached as Exhibit B, reports that peptides were prepared from various regions of the active C-terminal portion of GDF-8 (see, e.g., page 54, line 13, to page 55, line 12; see, also, FIG. 12), and used to immunize mice (see page 65, lines 23-28; see, also, page 62, line 27, to page 63, line 15), and that mice immunized with at least three of the peptides, including peptide 5 (pJS123; Group 6), peptide 17 (pJS129; Group 12), and peptide 18 (pJS130; Group 13), had statistically significant increased weight as compared to at least two of the three control groups (see paragraph bridging pages 67-68; see, also, FIG. 18). Further, when all the controls were grouped together and all of the immunized mice were grouped together, the immunized group showed a statistically significant increase in weight (*Id.*).

It is submitted that one skilled in the art, viewing the results of WO 99/42573, reasonably would have known that each of the different peptide fragments of GDF-8 likely would have induced different immune responses, including differences in the level of the immune response as well as in the specificity of the immune response. For example, the artisan would have known

that each of the peptides would correspond to different epitopes of the myostatin protein naturally expressed in the mice, and that the different epitopes would not likely generate exactly the same level and specificity of immune response. As such, it is submitted that animals immunized with different GDF-8 peptides would have had different antibody responses, which, in turn, would have resulted in varying levels of GDF-8 (myostatin) inhibition. Nevertheless, at least three of the Groups of immunized mice exhibited significantly increased weight as compared to the controls, and all of the immunized animals, when considered together, showed statistically significant increased weight gains over the controls. As such, it is submitted that WO 99/42573 suggests that, despite the amount of myostatin inhibition that occurred, all of the treated mice exhibited at least some increase in weight gain, which, it is submitted, is due at least in part to increased muscle mass.

Finally, Applicants point out that the claims require that the transgene, which comprises a nucleic acid sequence encoding a dominant negative Act RII operably linked to a regulatory element comprising a muscle specific promoter, "is expressed so as to result in elevated levels of the dominant negative Act RII and increased muscle mass (see claim 1), and further allow that the regulatory element can comprise, for example, an enhancer. In this respect, it is noted that muscle specific promoters are disclosed in the specification (see, e.g., paragraph 101, pages 37-38), or otherwise known in the art. For example, Marchand et al. (*Development* 117:947-959, 1993, a copy of which is attached as Exhibit C), describe a regulatory element of the desmin gene is a very strong promoter/enhancer that directs muscle-specific expression of a reporter gene (see Abstract). Further Klamut et al. (*Hum. Molec. Genet.* 5:1599-1606, 1996, a copy of which is attached as Exhibit D) describe an enhancer in muscle intron 1 of the dystrophin gene (see Abstract), and also note that enhancers have been identified in a number of muscle-specific genes, referring specifically to the muscle creatine kinase gene (see page 1600, left column, second full paragraph). Klamut et al. also note that the SV40 enhancer has been

reported to have activity in skeletal muscle similar to that of the myosin light chain 1/3 enhancer (see page 1603, paragraph bridging columns). As such, it is submitted that the skilled artisan, viewing the subject application, would have known of numerous regulatory elements comprising a muscle specific promoter useful for practicing the claimed invention.

Further, even if some experimentation may be required to determine, for example, whether expression of a dominant negative Act RII from a particular regulatory element comprising a muscle specific promoter (e.g., a creatine kinase promoter; see, e.g., paragraph 101, pages 37-38) or from such a regulatory element further comprising an enhancer (see, e.g., paragraph 144, page 55) is sufficient to obtain enhanced muscle mass, it is submitted that such experimentation would be routine and not require any inventive input, and would not amount to undue experimentation under the law (see, e.g., *In re Wands*, 8 USPQ2d 1400 (Fed. Cir. 1988), holding that, where direction and guidance, and working examples are provided, it was not undue experimentation to screen hybridoma; see section [2] at pages 1406-1407).

In summary, the specification discloses transgenic mice having variable levels of increased muscle mass, which can be due to variable expression of the transgene due to positional effects, and it was known in the art at the time the subject application was filed that expression of a dominant negative Act RII can inhibit gene expression in a dose-dependent manner, and that immunization of mice with various peptide fragments of GDF-8 results in increased weight of the mice. As such, it is submitted that one skilled in the art, viewing the specification and having knowledge of the art, would have known that dominant negative Act RII expression in muscle cells of a transgenic mammal can inhibit GDF-8 signal transduction in the muscle cells in a dose-dependent manner, and reasonably would have predicted that inhibition of GDF-8 signal transduction would correlate with muscle mass in the transgenic mammal. Further, the artisan would have known of numerous muscle-specific

regulatory elements having various levels of activity that could be used to drive expression of the dominant negative Act RII. Accordingly, it is submitted that undue experimentation would not have been required to practice the full scope of the claimed invention and, therefore, respectfully requested that the Examiner reconsider and withdraw the objection to the specification, and remove the corresponding rejection of the claims under 35 U.S.C. § 112, first paragraph.

In view of the amendments and the above remarks, it is submitted that the claims are in condition for allowance, and a notice to that effect is respectfully requested. The Examiner is invited to contact Applicants' undersigned representative if there are any questions relating to the subject application.

Enclosed is Check No. 564486 in the amount of $385.00 in payment of the request for continued examination fee. The Commissioner is hereby authorized to charge any other fees that may be associated with this communication, or credit any overpayment, to Deposit Account No. 50-1355.

Respectfully submitted,

Date: August 9, 2004

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Enclosures: Exhibits A, B, C and D